Amendments to the Claims

Claims 1-53 (Cancelled)

Claim 54 (Currently Amended): A type I polyketide synthase which produces a polyketide and which comprises a loading module and a plurality of extension modules, wherein:

a) said loading module ~~is adapted to~~ loads an optionally substituted malonyl and then ~~to~~ effects decarboxylation of the loaded ~~residue~~ moiety to provide a corresponding optionally substituted acetyl ~~residue~~ moiety for transfer to the first of said extension modules; ~~, wherein at least the first of said extension molecules is not naturally associated with a loading module that effects decarboxylation of an optionally substituted malonyl, and wherein~~

b) said loading module is of the form:

(engineered-KSq)-(AT)-(ACP), wherein:

i) ACP ~~represents~~ is an acyl carrier protein domain[[,]];

ii) AT ~~represents~~ is an acyltransferase domain which ~~is adapted to~~ loads an optionally substituted malonyl; and

iii) engineered-KSq ~~represents~~ is a domain which has been genetically engineered to effect decarboxylation of a loaded optionally substituted malonyl by mutating the active site cysteine residue to a glutamine residue; and

c) at least the first of said extension modules is not naturally associated with said loading module;

wherein the polyketide produced by the polyketide synthase is other than a 14-membered macrolide having a 13-methyl group due to incorporation of an unsubstituted acetate starter.

Claim 55 (Currently Amended): A type I polyketide synthase according to claim 54, wherein ~~the~~ said acyltransferase domain has an arginine residue in the active site.

Claim 56 (Currently Amended): A type I polyketide synthase according to claim 55, wherein ~~the~~ said acyltransferase domain is a natural extension module acyltransferase domain.

Claim 57 (Previously Presented): A type I polyketide synthase according to claim 54, wherein the engineered-KSq and acyltransferase domain pair produced by mutation occur together in an extension module in their unaltered state.

Claim 58 (Previously Presented): A type I polyketide synthase according to claim 55, wherein said acyltransferase domain is specific for loading with malonyl.

Claim 59 (Previously Presented): A type I polyketide synthase according to claim 55, wherein said acyltransferase domain is specific for loading with methylmalonyl.

Claim 60 (Currently Amended): A type I polyketide synthase according to claim 55, wherein said acyltransferase domain is ~~specific for loading with ethylmalonyl~~ selected from the group consisting of the acyltransferase domain of extension module 5 of the monensin polyketide synthase and the acyltransferase domain of extension module 5 of the spiramycin polyketide synthase.

Claim 61 (Currently Amended): A type I polyketide synthase according to claim 56, wherein said acyltransferase domain ~~corresponds to~~ is the acyltransferase of module 6 of the niddamycin polyketide synthase.

Claim 62 (Currently Amended): A type I polyketide synthase according to claim 56, wherein said acyltransferase domain ~~corresponds to~~ is the acyltransferase of module 4 of the FK506

polyketide synthase.

Claim 63 (Currently Amended): A type I polyketide synthase according to claim 54, wherein said polyketide synthase is ~~adapted~~ effective to synthesize a polyketide selected from

(a) 12- and 16-membered macrolides with acetate starter units;

(b) 12, 14, and 16-membered macrolides with propionate starter units;

(c) variants of rifamycin, avermectin, rapamycin, immunomycin and FK506 which differ from the natural compound in the incorporation of acetate starter units or propionate starter units; ~~or~~

(d) a polyketide wherein the starter unit ~~gave rise to a sidechain selected from allyl and hydroxymethyl~~ is derived from a loading domain comprising the acyltransferase domain of extension module 4 of the FK506 polyketide synthase; or

(e) a polyketide wherein the starter unit is derived from a loading domain comprising the acyltransferase domain of extension module 6 of the niddamycin polyketide synthase.

Claim 64 (New): A type I polyketide synthase which produces a polyketide and which comprises a loading module and a plurality of extension modules, wherein:

a) said loading module loads an optionally substituted malonyl and then effects decarboxylation of the loaded moiety to provide a corresponding optionally substituted acetyl moiety for transfer to the first of said extension modules;

b) said loading module is of the form:

(KSq)-(AT)-(ACP), wherein:

i) ACP is an acyl carrier protein domain;

ii) AT is an acyltransferase domain which loads an optionally substituted malonyl and is selected from the group consisting of the acyltransferase domain of module 6 of the

niddamycin polyketide synthase, the acyltransferase domain of module 4 of the FK506 polyketide synthase, the acyltransferase domain of module 2 of the rapamycin polyketide synthase; the acyltransferase domain of module 5 of the spiramycin polyketide synthase, and the acyltransferase domain of module 5 of the monensin polyketide synthase; and

iii) KSq is a domain which effects decarboxylation of a loaded optionally substituted malonyl and which differs from a ketosynthase domain of an extension module by having a glutamine residue in place of the cysteine residue in the active site; and

c) at least the first of said extension modules is not naturally associated with said loading module;

wherein the polyketide produced by the polyketide synthase is other than a 14-membered macrolide having a 13-methyl group due to incorporation of an unsubstituted acetate starter.

Claim 65 (New): The type I polyketide synthase according to claim 64, wherein (AT) is the acyltransferase domain of module 6 of the niddamycin polyketide synthase,

Claim 66 (New): The type I polyketide synthase according to claim 64, wherein (AT) is the acyltransferase domain of module 4 of the FK506 polyketide synthase.

Claim 67 (New): The type I polyketide synthase according to claim 64, wherein (AT) is the acyltransferase domain of module 2 of the rapamycin polyketide synthase.

Claim 68 (New): The type I polyketide synthase according to claim 64, wherein (AT) is the acyltransferase domain of module 5 of the spiramycin polyketide synthase.

Claim 69 (New): The type I polyketide synthase according to claim 64, wherein (AT) is the acyltransferase domain of module 5 of the monensin polyketide synthase.

Claim 70 (New): The type I polyketide synthase according to claim 64, wherein the KSq domain is the KSq domain of the oleandomycin loading module.

Claim 71 (New): The type I polyketide synthase according to claim 64, wherein said extension modules are selected from the group consisting of extension modules from the erythromycin, rifamycin, avermectin, rapamycin, immunomycin, or FK506 polyketide synthases.

Claim 72 (New): A type I polyketide synthase which produces a polyketide and which comprises a loading module and a plurality of extension modules, wherein:

a) said loading module loads an optionally substituted malonyl and then effects decarboxylation of the loaded moiety to provide a corresponding optionally substituted acetyl moiety for transfer to the first of said extension modules;

b) said loading module is of the form:

(KSq)-(AT)-(ACP), wherein:

i) ACP is an acyl carrier protein domain;

ii) AT is an acyltransferase domain which loads an optionally substituted malonyl; and

iii) KSq is the KSq domain of the oleandomycin loading module; and

c) at least the first of said extension modules is not naturally associated with said loading module;

wherein the polyketide produced by the polyketide synthase is other than a 14-membered macrolide having a 13-methyl group due to incorporation of an unsubstituted acetate starter.

Claim 73 (New): A type I polyketide synthase which produces a polyketide and which comprises a loading module and a plurality of extension modules, wherein:

a) said loading module loads an optionally substituted malonyl and then effects decarboxylation of the loaded moiety to provide a corresponding optionally substituted acetyl moiety for transfer to the first of said extension modules;

b) said loading module is of the form:

(KSq)-(AT)-(ACP), wherein:

i) ACP is an acyl carrier protein domain;

ii) AT is an acyltransferase domain which loads an optionally substituted malonyl; and

iii) KSq is a domain which effects decarboxylation of a loaded optionally substituted malonyl and which differs from a ketosynthase domain of an extension module by having a glutamine residue in place of the cysteine residue in the active site;

c) at least the first of said extension modules is not naturally associated with said loading module; and

d) said loading module is the loading module of the monensin polyketide synthase;

wherein the polyketide produced by the polyketide synthase is other than a 14-membered macrolide having a 13-methyl group due to incorporation of an unsubstituted acetate starter.

Claim 74 (New): A type I polyketide synthase which produces a polyketide and which comprises a loading module and a plurality of extension modules, wherein:

a) said loading module loads an optionally substituted malonyl and then effects decarboxylation of the loaded moiety to provide a corresponding optionally substituted acetyl moiety for transfer to the first of said extension modules;

b) said loading module is of the form:

(KSq)-(AT)-(ACP), wherein:

i) ACP is an acyl carrier protein domain;

ii) AT is an acyltransferase domain which loads an optionally substituted malonyl; and

iii) KSq is a domain which effects decarboxylation of a loaded optionally substituted malonyl and which differs from a ketosynthase domain of an extension module by having a glutamine residue in place of the cysteine residue in the active site;

c) at least the first of said extension modules is not naturally associated with said loading module; and

d) said loading module is the loading module of the oleandomycin polyketide synthase;

wherein the polyketide produced by the polyketide synthase is other than a 14-membered macrolide having a 13-methyl group due to incorporation of an unsubstituted acetate starter.

Claim 75 (New): A type I polyketide synthase which produces a 12- or 14- membered macrolide and which comprises a loading module and a plurality of extension modules, wherein:

a) said loading module loads an optionally substituted malonyl and then effects decarboxylation of the loaded moiety to provide a corresponding optionally substituted acetyl moiety for transfer to the first of said extension modules;

b) said loading module is of the form:

(KSq)-(AT)-(ACP), wherein:

i) ACP is an acyl carrier protein domain;

ii) AT is an acyltransferase domain which loads an optionally substituted malonyl; and

iii) KSq is a domain which effects decarboxylation of a loaded optionally substituted malonyl and which differs from a ketosynthase domain of an extension module by having a

glutamine residue in place of the cysteine residue in the active site;

c) at least the first of said extension modules is not naturally associated with said loading module; and

d) said loading module is the loading module of the tylosin polyketide synthase;

wherein the polyketide produced by the polyketide synthase is other than a 14-membered macrolide having a 13-methyl group due to incorporation of an unsubstituted acetate starter.

Claim 76 (New): A type I polyketide synthase which produces a polyketide and which comprises a loading module and a plurality of extension modules, wherein:

a) said loading module loads an optionally substituted malonyl and then effects decarboxylation of the loaded moiety to provide a corresponding optionally substituted acetyl moiety for transfer to the first of said extension modules;

b) said loading module is of the form:

(KSq)-(AT)-(ACP), wherein:

i) ACP is an acyl carrier protein domain;

ii) AT is an acyltransferase domain which loads an optionally substituted malonyl; and

iii) KSq is a domain which effects decarboxylation of a loaded optionally substituted malonyl and which differs from a ketosynthase domain of an extension module by having a glutamine residue in place of the cysteine residue in the active site;

c) at least the first of said extension modules is not naturally associated with said loading module; and

d) said loading module is the loading module of the spiramycin polyketide synthase;

wherein the polyketide produced by the polyketide synthase is

other than a 14-membered macrolide having a 13-methyl group due to incorporation of an unsubstituted acetate starter.